

Managing Soil Microorganisms to Improve Productivity of Agrosoil Ecosystems

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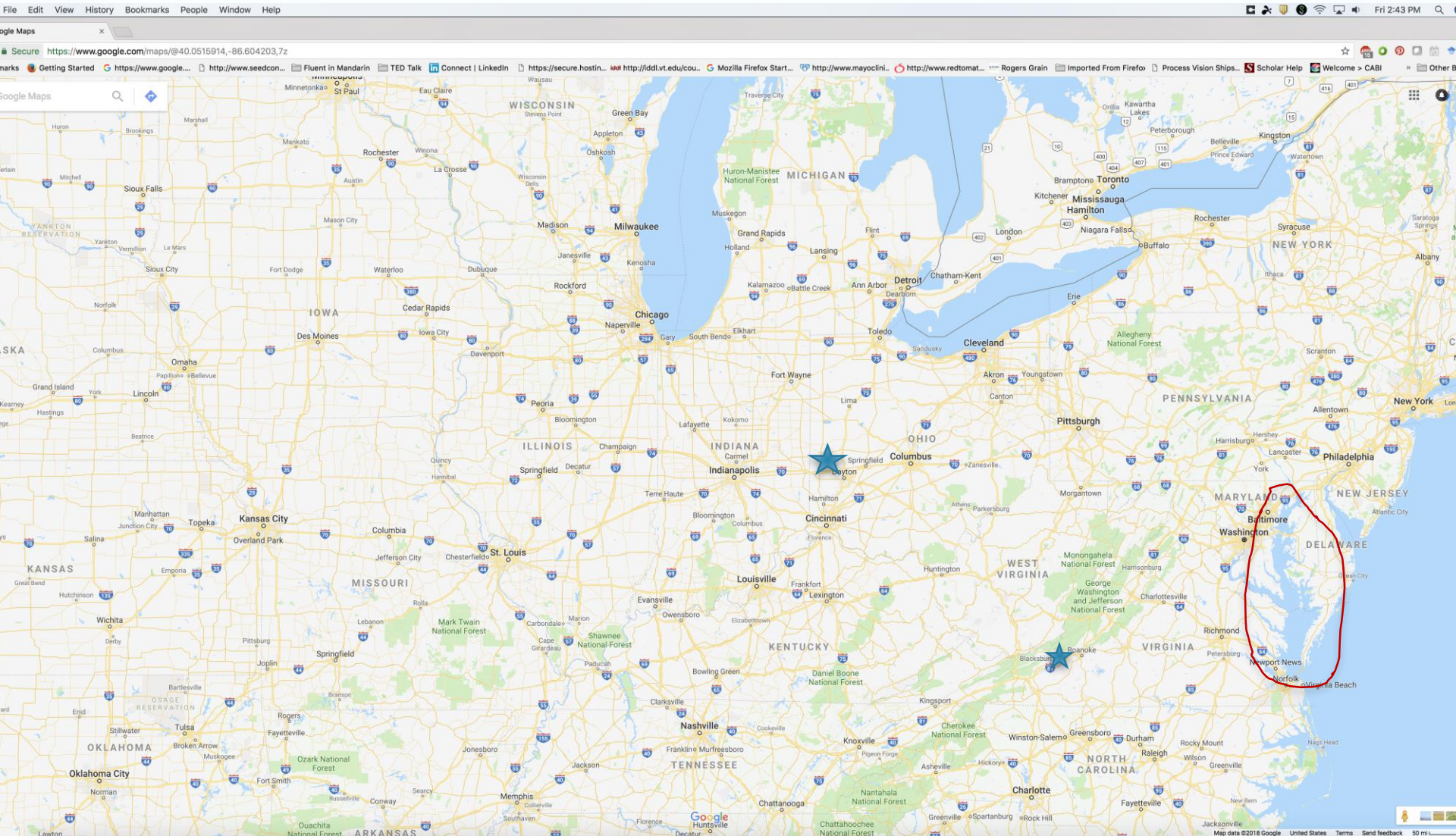
Thermogradient Systems, LLC Blacksburg, VA

Virginia Tech Corporate Research Center, Blacksburg, VA



My Farm

(Newton Township, Miami Co. West Central Ohio)





(Some) Challenges for Modern Farmers

- Low commodity prices and decreased profitability caused by higher production costs
- Climate Change (more rain as well as drought)
- Environmental impact of farming and particularly nutrient management

Consequences of Global Climate Change

- Greater variability
- More frequent severe weather
- Drought in some areas but more rainfall and flooding in other areas (like the southern Great Lakes)

Dealing with Seasonal Flooding

Data shows more frequent rainfall events of 2" or more at one time over the past 20 years in the Great Lakes Region of the US



Artificial drainage to reduce plant disease and increase productivity



Installing subsurface tile drainage system in crop fields



The drainage to improve crop production



Family Farm Miami Co. Ohio since 1803

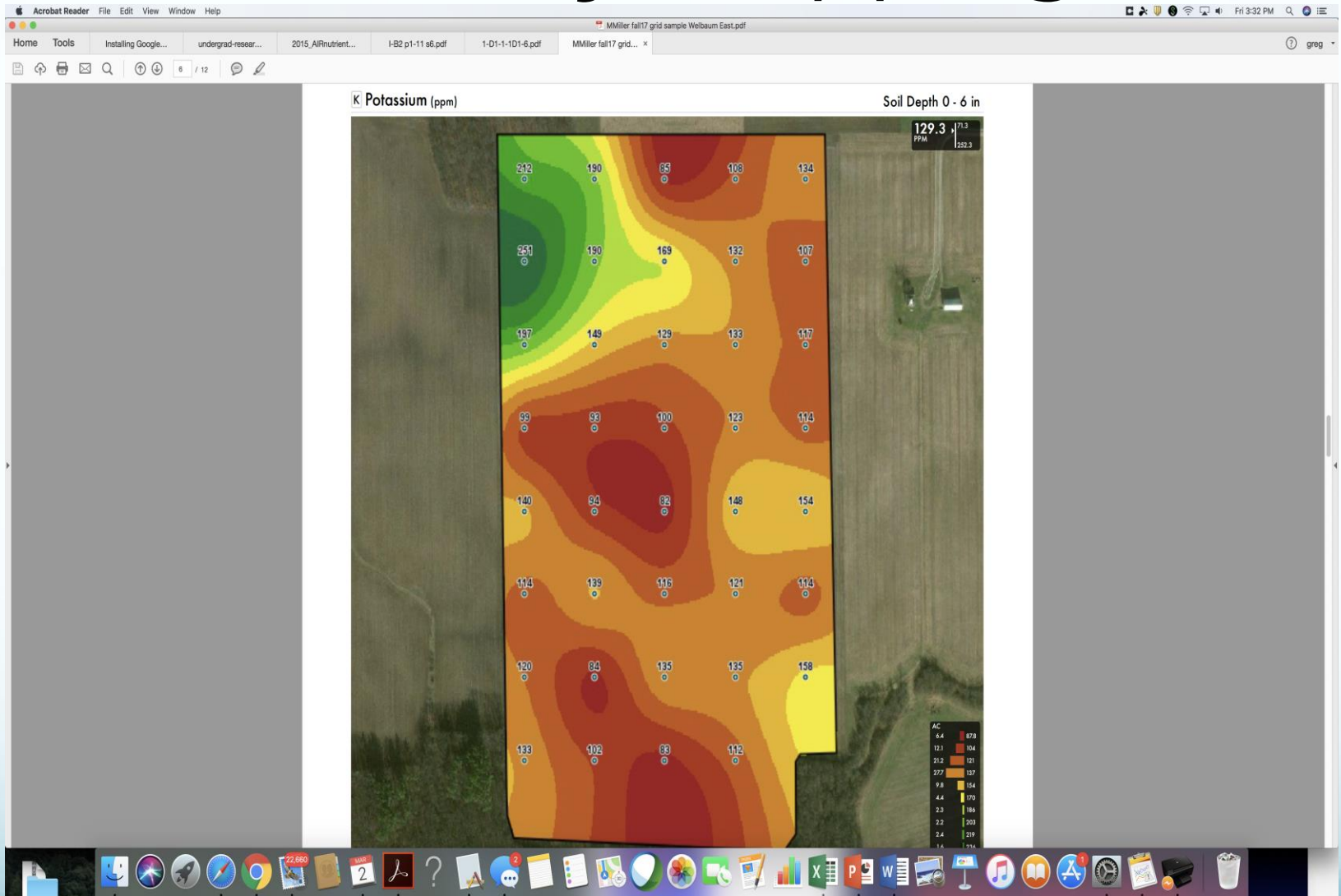
Picture taken by drone



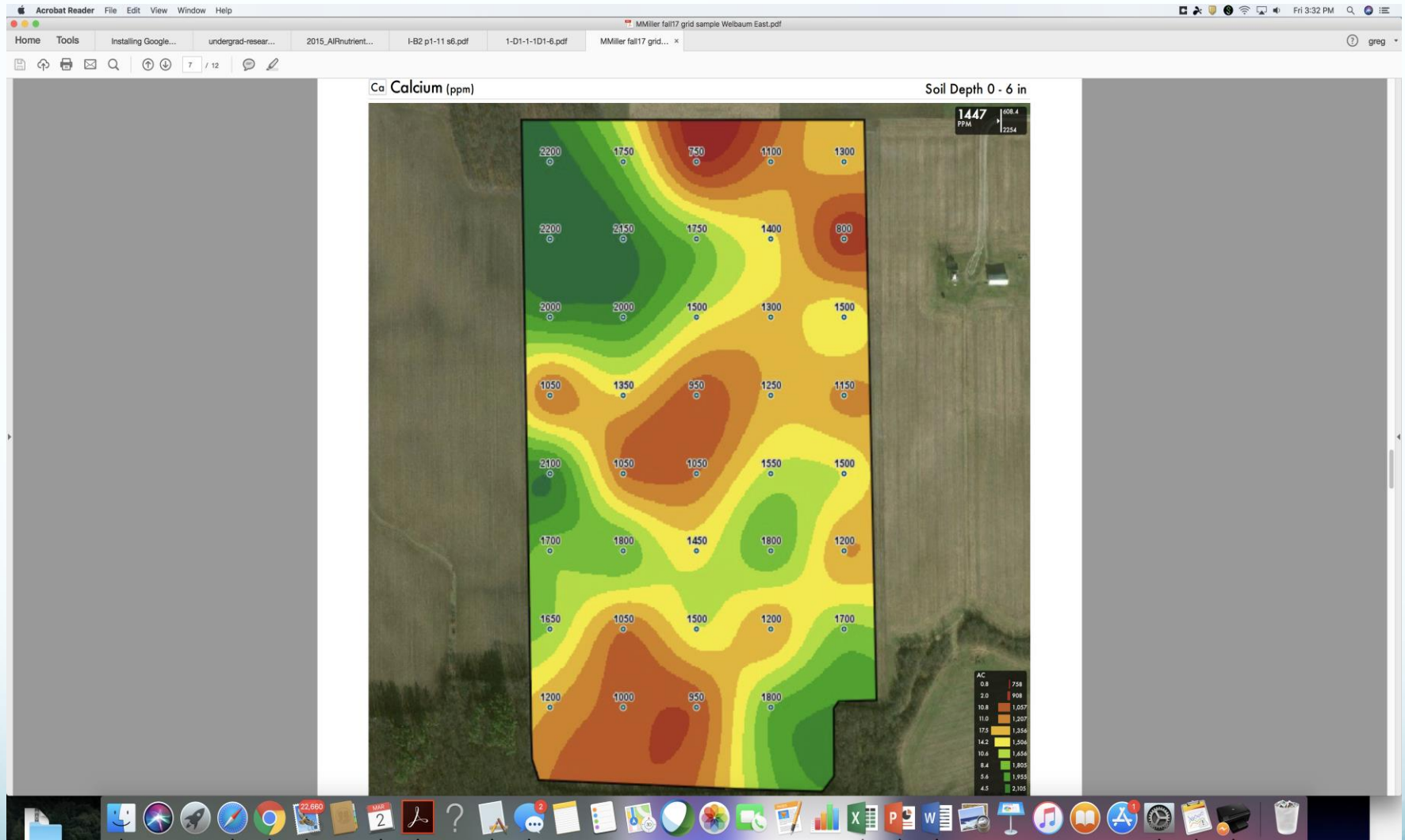
Downside to field drainage systems?

- Our system worked very effectively to remove excess rainfall! Installation costs \$80,000/100 acres
- Do drainage systems contribute to increased runoff and nutrient loss?
- Studies suggest no but nutrient loss is definitely possible with ill-timed applications coupled with excessive moisture events
- Applying the minimum amount of nutrients required using fertility mapping is one approach to reducing nutrient waste/pollution.

Fertility Mapping



Fertility Mapping



Fertility Mapping



CPS - Greenville OH 3021
415 South Ohio Street

Office 937-548-1816

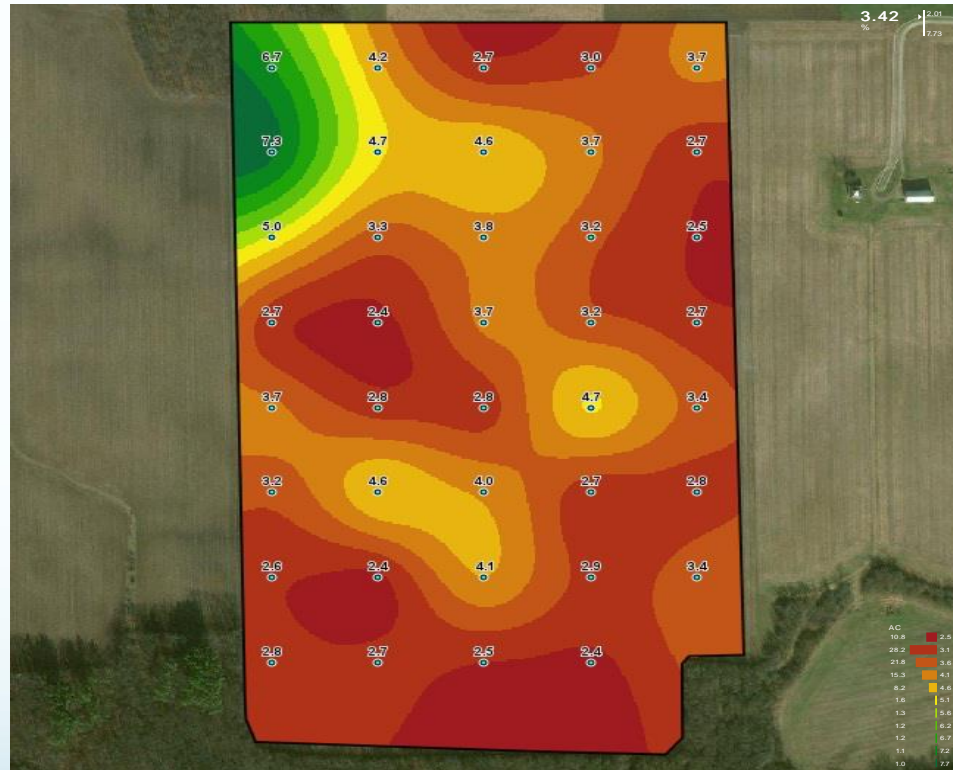
Lab Results Map

Soil Sample
2017-10-13

Grower Field	MARK MILLER Welbaum East	Farm Area (acre)	MILLER Mark 91.50	Lab	A&L Great Lakes
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OM Organic Matter (%)

Soil Depth 0 - 6 in



Fertility Mapping



CPS - Greenville OH 3021
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Lab Results Map

Soil Sample
2017-10-05

Grower
Field

MARK MILLER
Welbaum West

Farm
Area (acre)

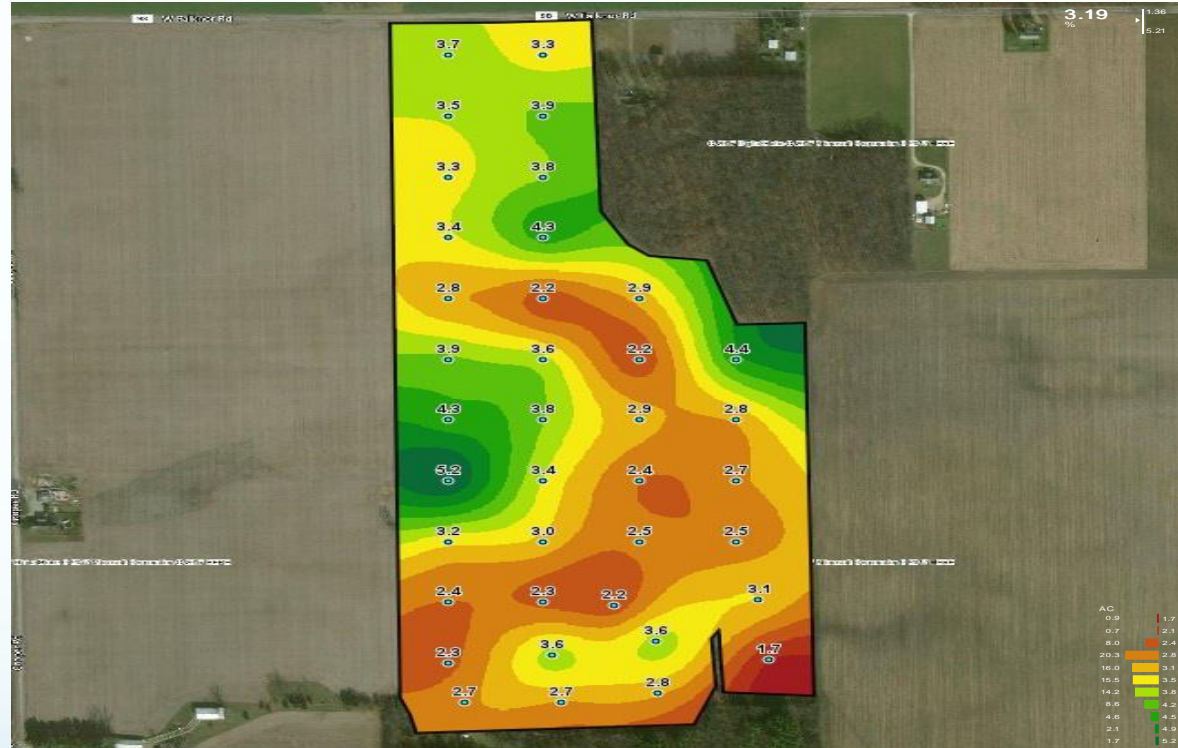
MILLER Mark
92.47

Lab

A&L Great Lakes

OM Organic Matter (%)

Soil Depth 0 - 6 in

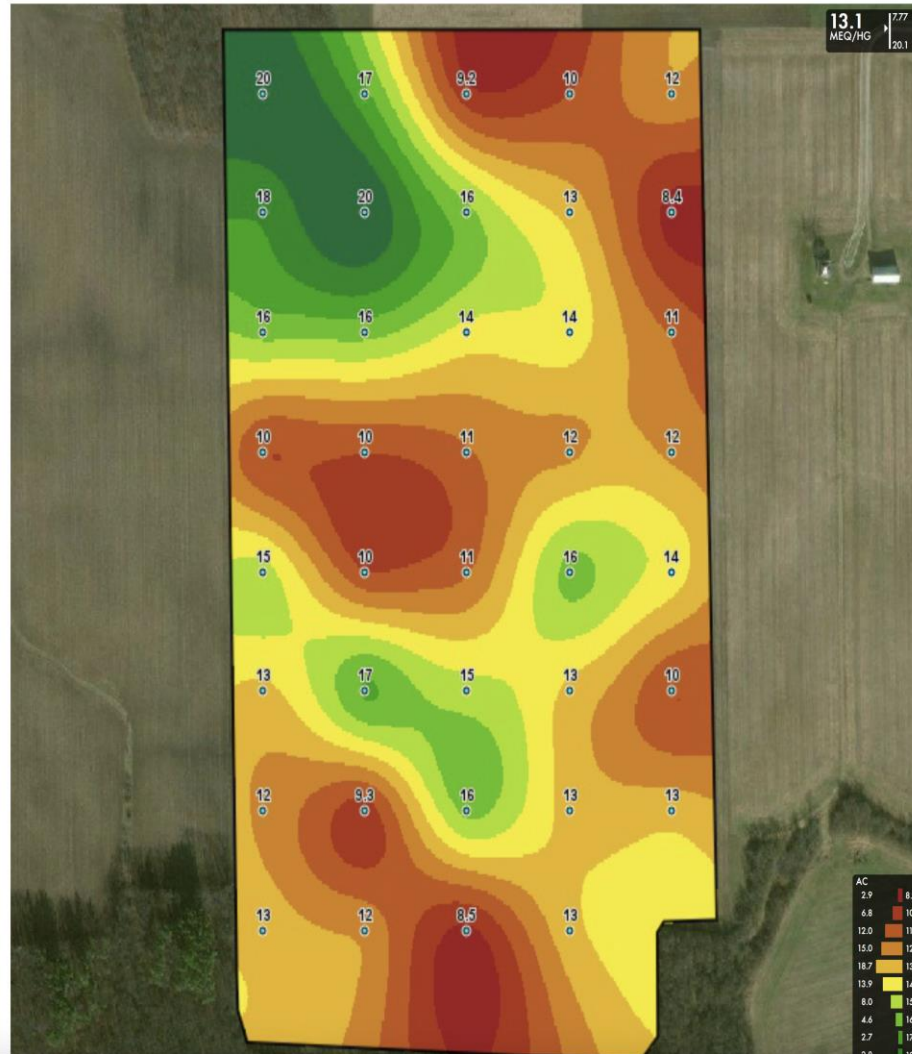


Fertility Mapping



CEC Cation Exchange Capacity (meq/hg)

Soil Depth 0 - 6 in



Fertility Mapping

- Allows variable rate fertilizer applications which should save inputs.
- Allows variable rate seeding to tailor plant populations to soil fertility.
- Enables comparisons of soil inputs to yield data to identify other production problems.
- Accurate GPS field/fertility mapping will lead to autonomous planting and harvesting operations.

Compensating for Reduced Organic Matter with Soil Priming

- What is soil priming?
- **Priming** or a "Priming Effect" is said to occur when something that is added to **soil** or compost affects the rate of decomposition occurring on the **soil** organic matter (SOM), either positively or negatively.
- Soils can be primed by planting certain types of cover crops or by adding amendments to soil.
- My work has involved using disaccharides to soils to increase microbial activity

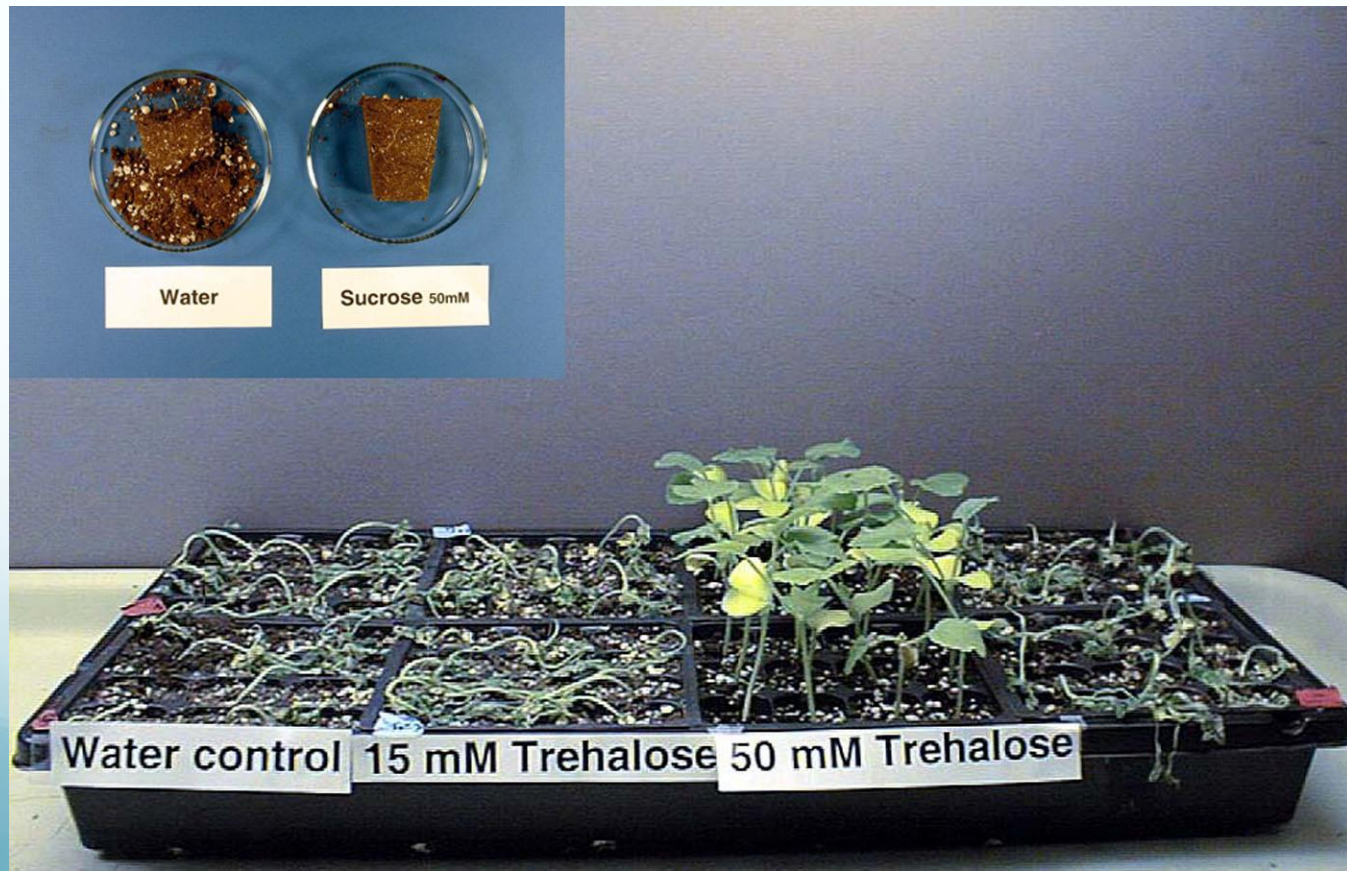
Soil Priming (to improve soil characteristics for the next crop)

1. Cover crops
2. Direct inputs of beneficial compounds

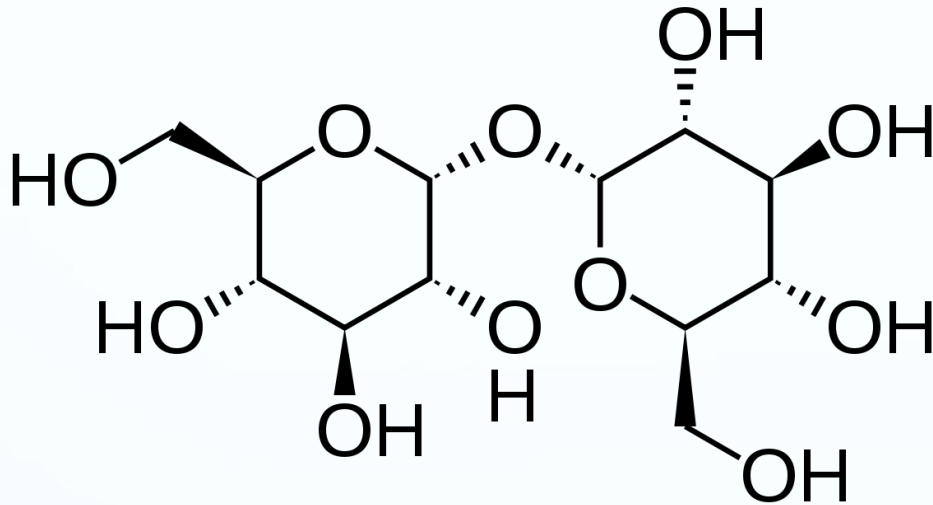


Priming with Carbon Sources to Feed Soil Microbes

Cantaloupe transplants growing in Sunshine Mix I,
coarse sphagnum moss and perlite
(Fissons, Vancouver, BC, Canada)



Trehalose



A disaccharide formed by an α , α -1,1-glucoside bond between two α -glucose units

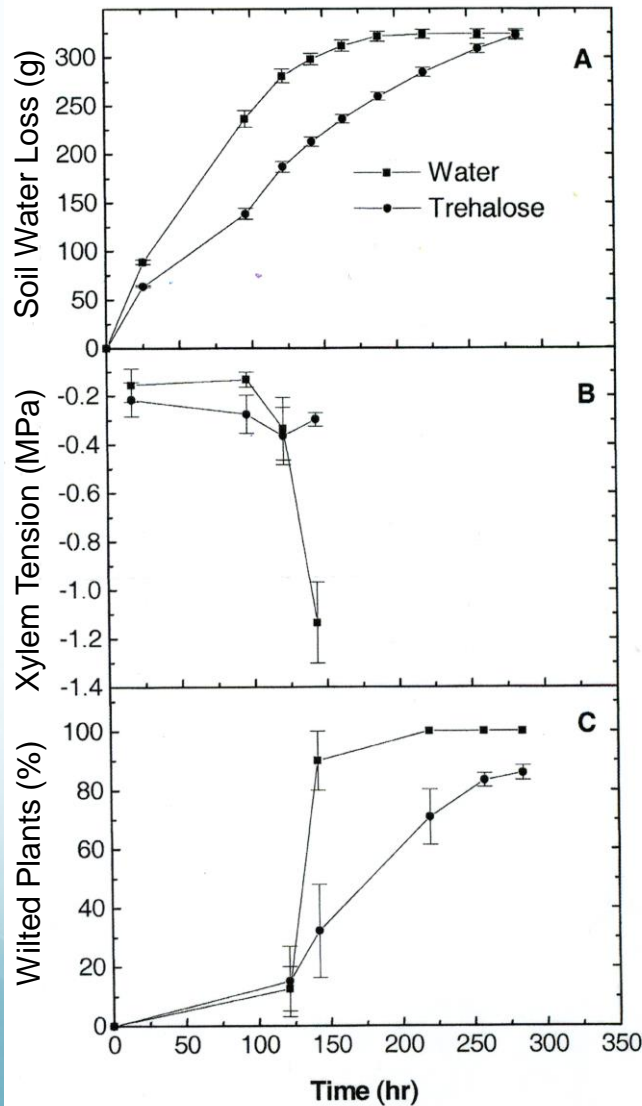


Water



Sucrose 50mM

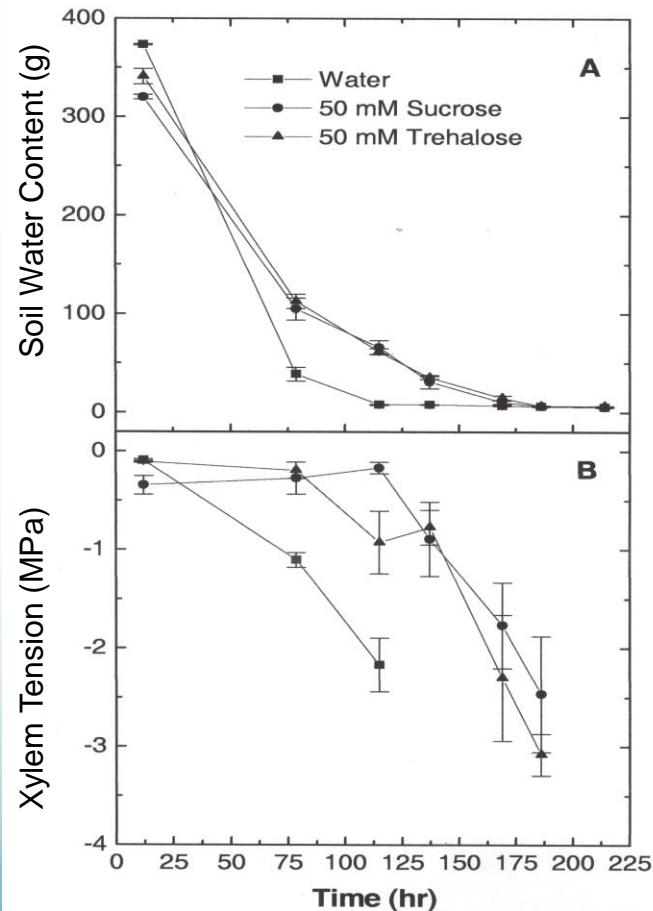
Soil water loss and seedling water relations following trehalose treatment



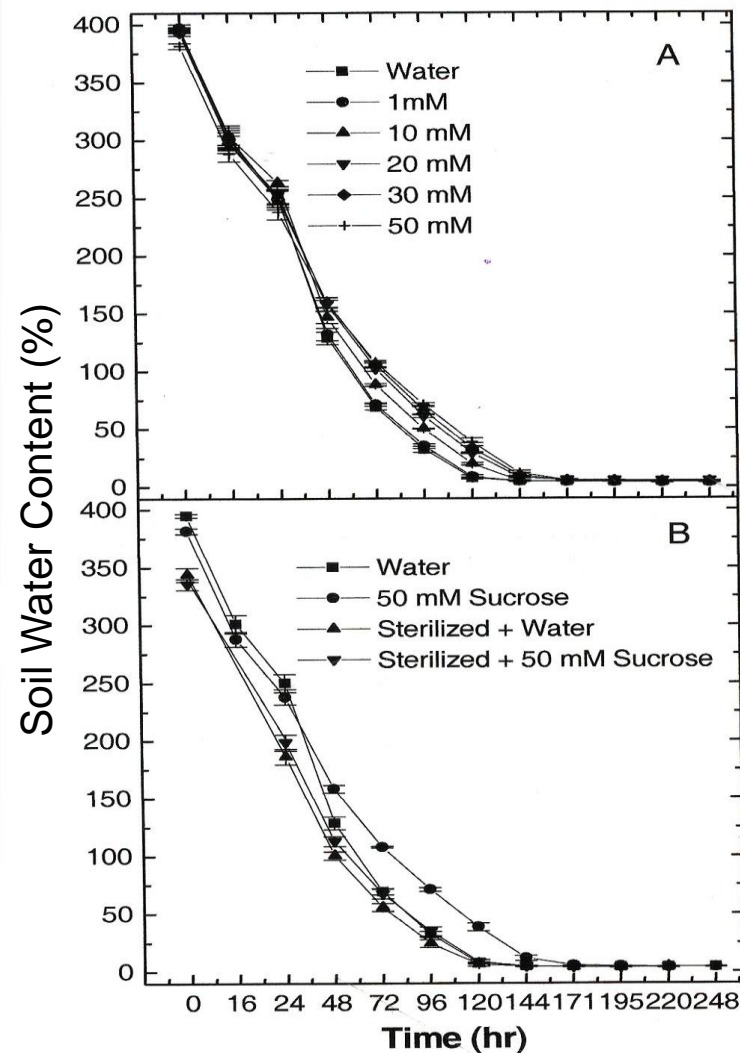
Cumulative water loss from plug trays filled with equal volumes of Sunshine Mix potting soil primed with 20 mL of 50 mM trehalose or water in a growth chamber. Error bars represent \pm SE of four replications of 12 plants each for each time interval (A). Sequential muskmelon seedling water potentials measured with a pressure bomb after an initial application with 20 mL of 50 mM trehalose or water (B). Development of wilting symptoms after 20 mL solutions of water or 50 mM trehalose were applied to individual tray cells (C). Error bars represent \pm SE of four replications of 12 plants each.

Changes in potting mix soil moisture content with priming

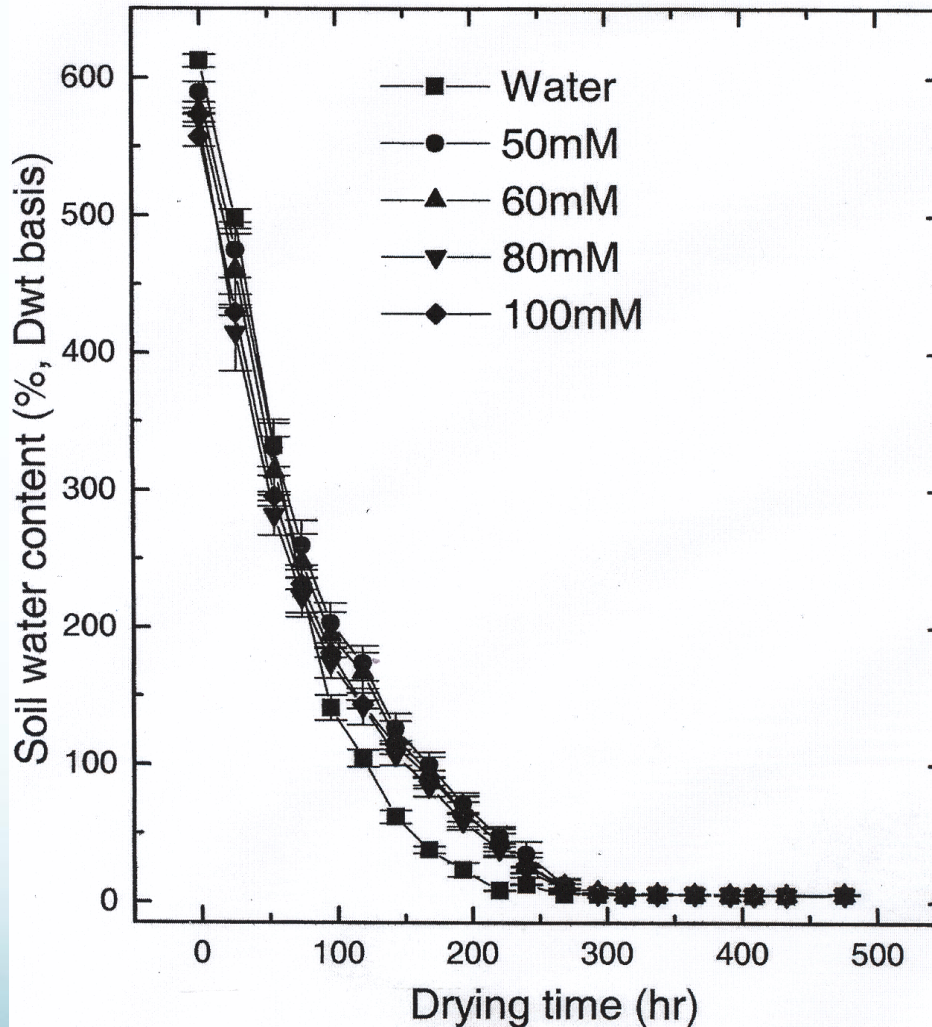
Changes in potting-mix moisture content following priming with 8 mL of water, 50 mM sucrose, or 50 mM trehalose. Potting-mix moisture contents were determined from fresh and oven dry weights of media from 4, 12-celled trays (A). Pressure-bomb measurements of 10-day-old seedling water potentials from destructively harvested plants grown in potting mix primed with sucrose or trehalose four days after seedling emergence. Error bars represent \pm SE of four replications at each time point (B).



Changes in Sunshine Mix Moisture Content With Different Treatments



Additions of greater than 50mM sucrose have no effect on soil water content



Pressure plate determination of moisture content

Table 1. Sunshine growing media water content after treatment with water or 50 mM sucrose and equilibration at 0.4 or 1.5 MPa on a pressure plate. Values represent 3 replications of each treatment. Water content was calculated after oven drying at 98°C.

Soil Treatment	Equilibration Pressure	
	0.4 MPa	1.5 MPa
Water content		
g kg ⁻¹		
Water	205 b	155 b
50 mM Sucrose	338 a	278 a
LSD	88	34

Effects of sucrose on other soil types

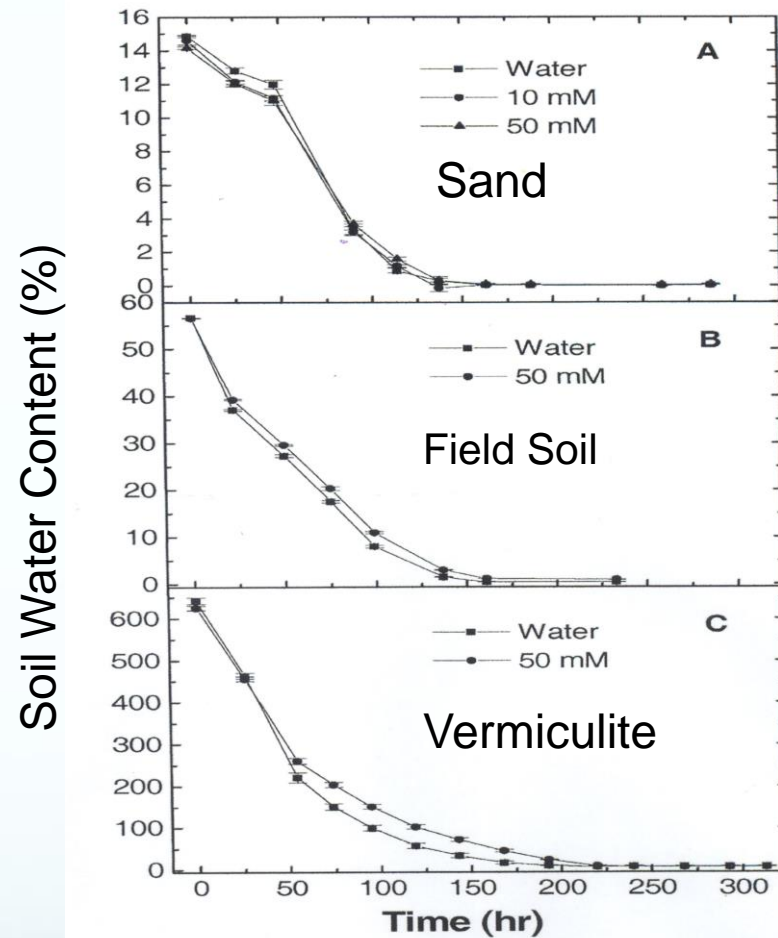
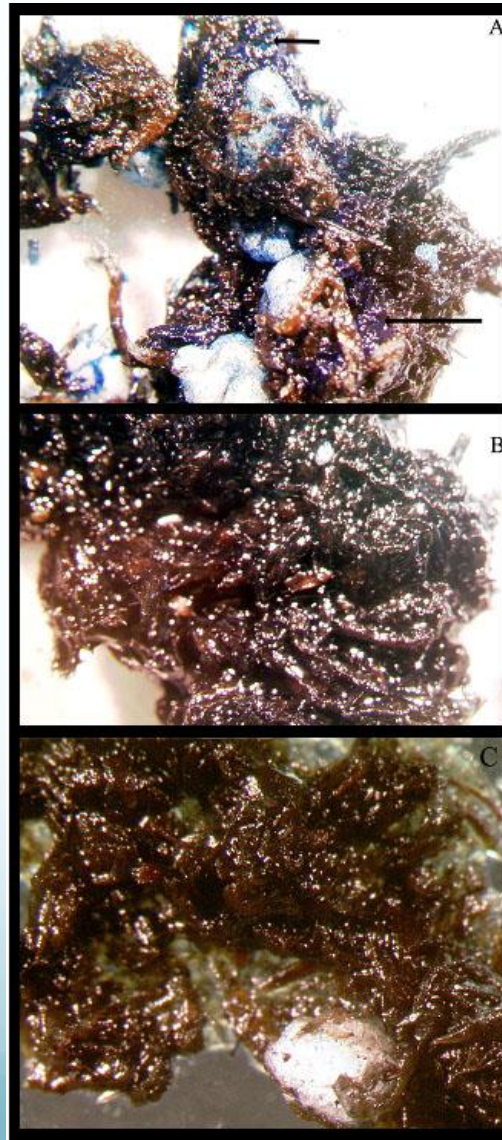
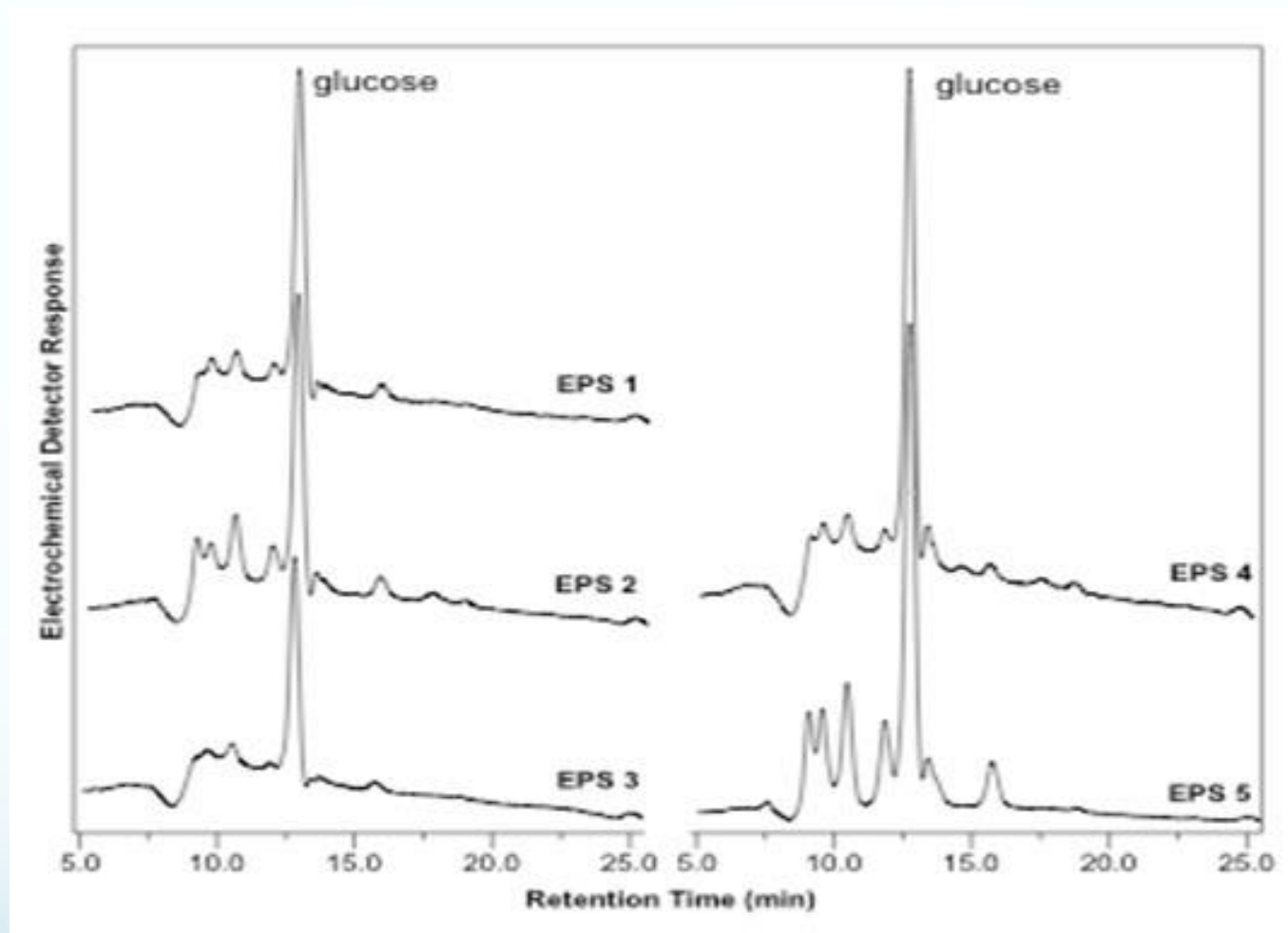


Figure 5. Comparison of the water contents of acid-washed **sand** in a growth chamber after a single treatment of 8 mL of water, 10 or 50 mM sucrose (A) **Hayter clay loam field soil** treated with water or 50 mM sucrose (B) or finely ground **vermiculate rooting media** treated with water or 50 mM sucrose (C).

Alcian blue staining after treatment with sucrose



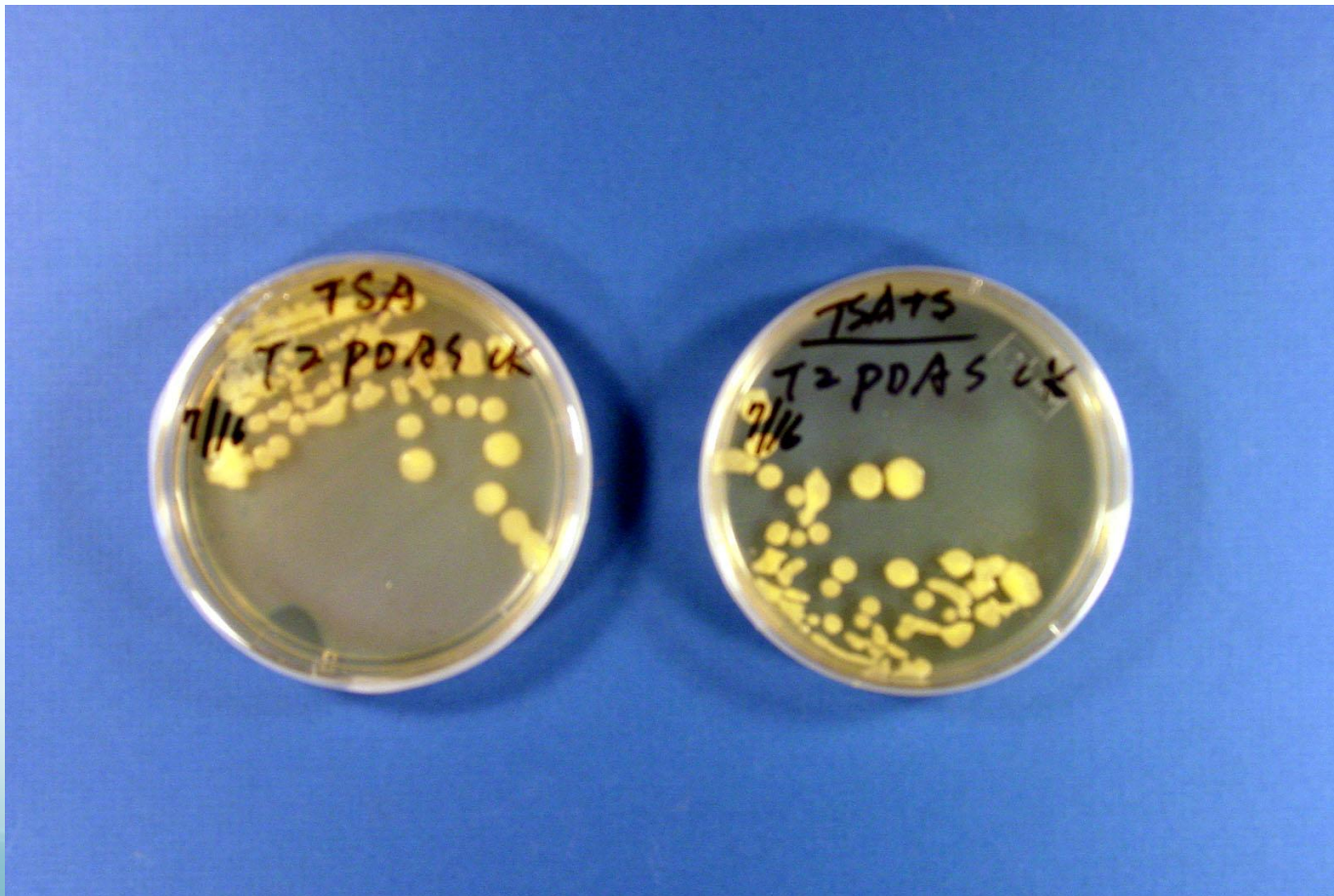
HPLC analysis of sugar induced polymer



Chromatograms of neutral monosaccharides present in the EPS samples. Glucose is the predominant monosaccharide, comprising between 55-65% of the total neutral sugars. Variability in the levels of the other monosaccharides prevented accurate quantification.

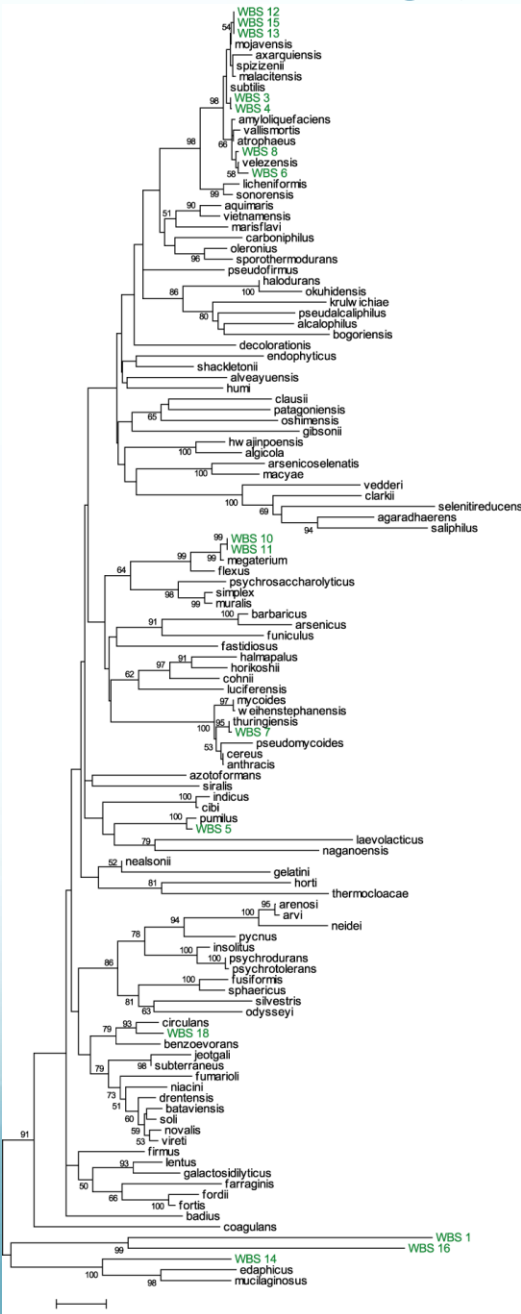
Colonies of bacteria were plated from sucrose treated soil using TSA media

Some bacteria may not have been culturable



Colony Sequence Identities

Sixteen bacterial colonies isolated from 50 mM sucrose-treated Sunshine Potting Mix with enhanced moisture retention were genetically identified based on 16S rRNA sequence homology. The neighbor-joining tree shows interspecies relationships among sample matches. Distances can be estimated by summing horizontal differences using the N-join distance key at the bottom of the diagram.

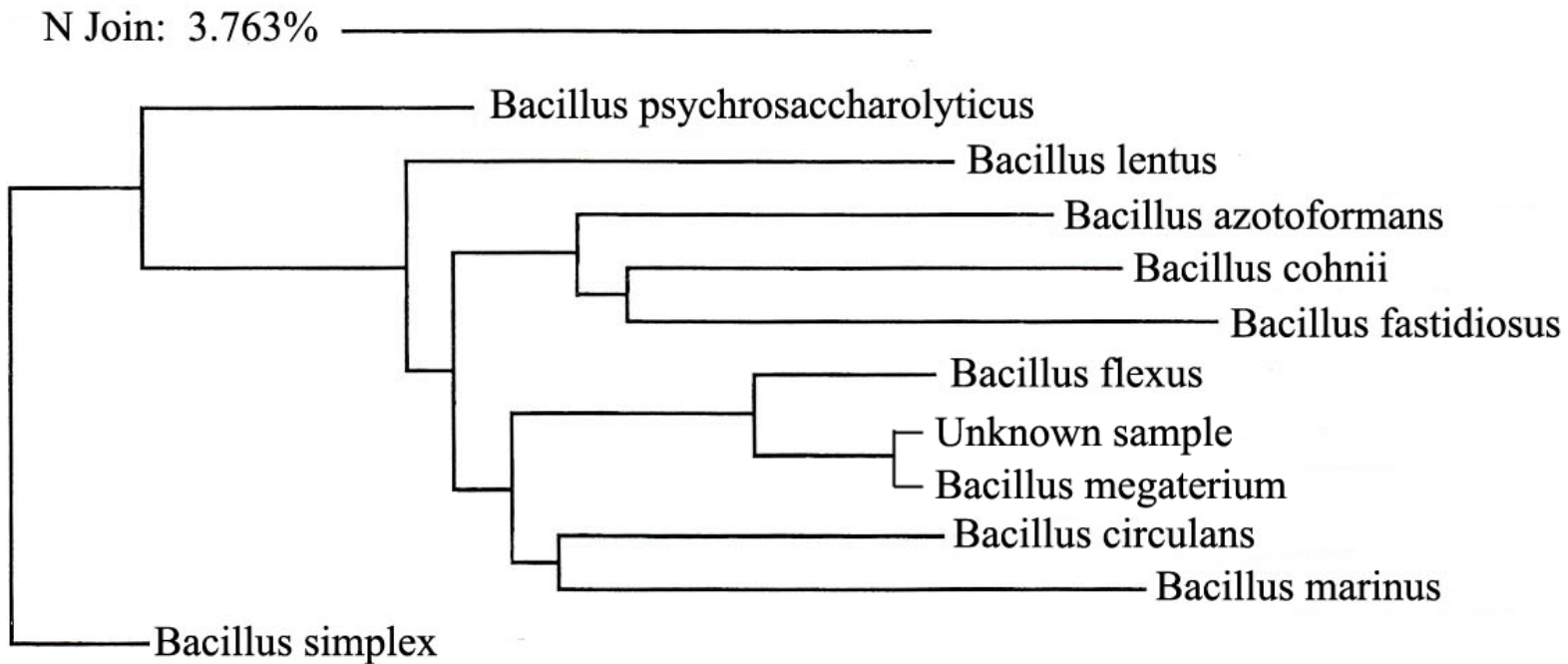


Putative colony identity based on 16S sequencing

Table 2. Bacterial colonies isolated from potting mix and cultured in TSA enriched with 50 mM sucrose. 16S rRNA amplification products were sequenced and database matches are listed below. Relationships among bacteria are summarized in Fig. 8

Sample	Gram Stain	Appearance	16s Analysis	Match
WBS1	Unknown	Unknown	yes	<i>Curtobacterium pusillum</i>
WBS2	pos	rod	yes	<i>Bacillus subtilis subtilis</i>
WBS3	pos	rod	yes	<i>Bacillus subtilis subtilis</i>
WBS4	pos	rod	yes	<i>Bacillus subtilis subtilis</i>
WBS5	pos	rod	yes	<i>Bacillus pumilus</i>
WBS6	pos	rod	yes	<i>Bacillus amyloliquefaciens</i>
WBS7	pos	rod	yes	<i>Bacillus thuringiensis</i>
WBS8	pos	rod	yes	<i>Bacillus amyloliquefaciens</i>
WBS10	pos	rod	yes	<i>Bacillus megaterium</i>
WBS11	pos	rod	yes	<i>Bacillus megaterium</i>
WBS12	pos	rod	yes	<i>Bacillus mojavenensis</i>
WBS13	pos	rod	yes	<i>Bacillus mojavenensis</i>
WBS14	Unknown	Unknown	yes	<i>Paenibacillus lautus</i>
WBS15	Unknown	Unknown	yes	<i>Bacillus mojavenensis</i>
WBS16	Unknown	Unknown	yes	<i>Brevundimonas sp.</i>
WBS17	Unknown	Unknown	no growth	
WBS18	Unknown	Unknown	yes	<i>Bacillus circulans</i>

Identity of the unknown bacterium



Conclusions

- The natural biofilm discovered in this study improved soil moisture holding characteristics
- A natural biofilm was produced in the commercial sphagnum moss-based potting mix used to grow melon seedlings. The source of the sphagnum used was a bog in New Brunswick, Provenance Canada
- The biofilm was most likely the result of a consortium of bacteria, both culturable and nonculturable, adapted to the specific environment of the potting mix.
- Attempts to consistently produce the biofilm in other soil types were unsuccessful.

Conclusions

- More research on soil microbiome may lead to discovery of biofilm producers for different environments and soil types. This is an active field of research in the US.
- The biofilm we discovered was a glucose-based polymer. Similar polymers could be produced and added as soil amendments to improve soil quality.
- Adding carbon sources to soils in small quantities as fertilizer can stimulate microbial activity which may benefit soil health and crop production particularly in situations where intensive farming has depleted organic matter. Farmers should consider the nutritional needs of soil microbes as well as crop plants.

Priming Soil References

- Welbaum, G. E., Shen, Z. X., Watkinson, J. I., Wang, C. L., & Nowak, J. (2009). **Priming soilless growing medium with disaccharides stimulated microbial biofilm formation, and increased particle aggregation and moisture retention during muskmelon transplant production.** Journal of the American Society for Horticultural Science, 134(3), 387-395.
- Welbaum, Gregory E. , Sturz, Antony V. , Dong, Zhongmin and Nowak, Jerzy (2004) '**Managing Soil Microorganisms to Improve Productivity of Agro-Ecosystems**', **Critical Reviews in Plant Sciences**", 23: 2, 175 — 193

Concerns with Current Traditional Fertilizer Application Procedures

- Fertilizer applications
 - Commercial 3rd party application (who test and apply fertilizers)
 - Timing coincides with the applicators schedule and not best use practices e.g. fertilizer applied the fall before spring planting
 - Too much reliance on N stabilizers like N-butyl thiophosphoric triamine
- Consequence:
 - Too much fertilizer is being wasted, costing farmer's money, and becoming a pollutant

Nutrient Management Reports

(now required in some environmentally sensitive regions of the US)

Two nutrient reports are now required by the state of Maryland for growers in the Chesapeake Bay water shed

- Nutrient Management Annual Implementation Report
- Nutrient Management Plan
 - INCOMPLETE INFORMATION WILL BE DEEMED INADEQUATE AND MAY FORCE MDA TO SEEK MONETARY PENALTIES.
- Manure or Organics Applied
- Fertilizers applied for each crop grown

Nutrient Application Plans Must Include the Following

- NUTRIENT APPLICATION REQUIREMENTS
 - (1) Setbacks for Nutrient Application,
 - (2) Application Timing for all nutrients, organic and chemical
 - (3) Temporary Field Stockpiling (staging) of Organic Materials.
 - Prescribed amounts of nutrients to apply for each crop grown

Nutrient Application Plan Considerations

- The plan must describe how the application of nutrients will vary depending on the crop, season, nutrient source, and weather conditions.
- The plan must follow “Nutrient Application Requirements,” to maximize plant utilization efficiency as described in regulations for each state or region
- Application plan procedures must minimize the potential for nutrient movement to sensitive areas and losses to surrounding water bodies, including surface and groundwater.

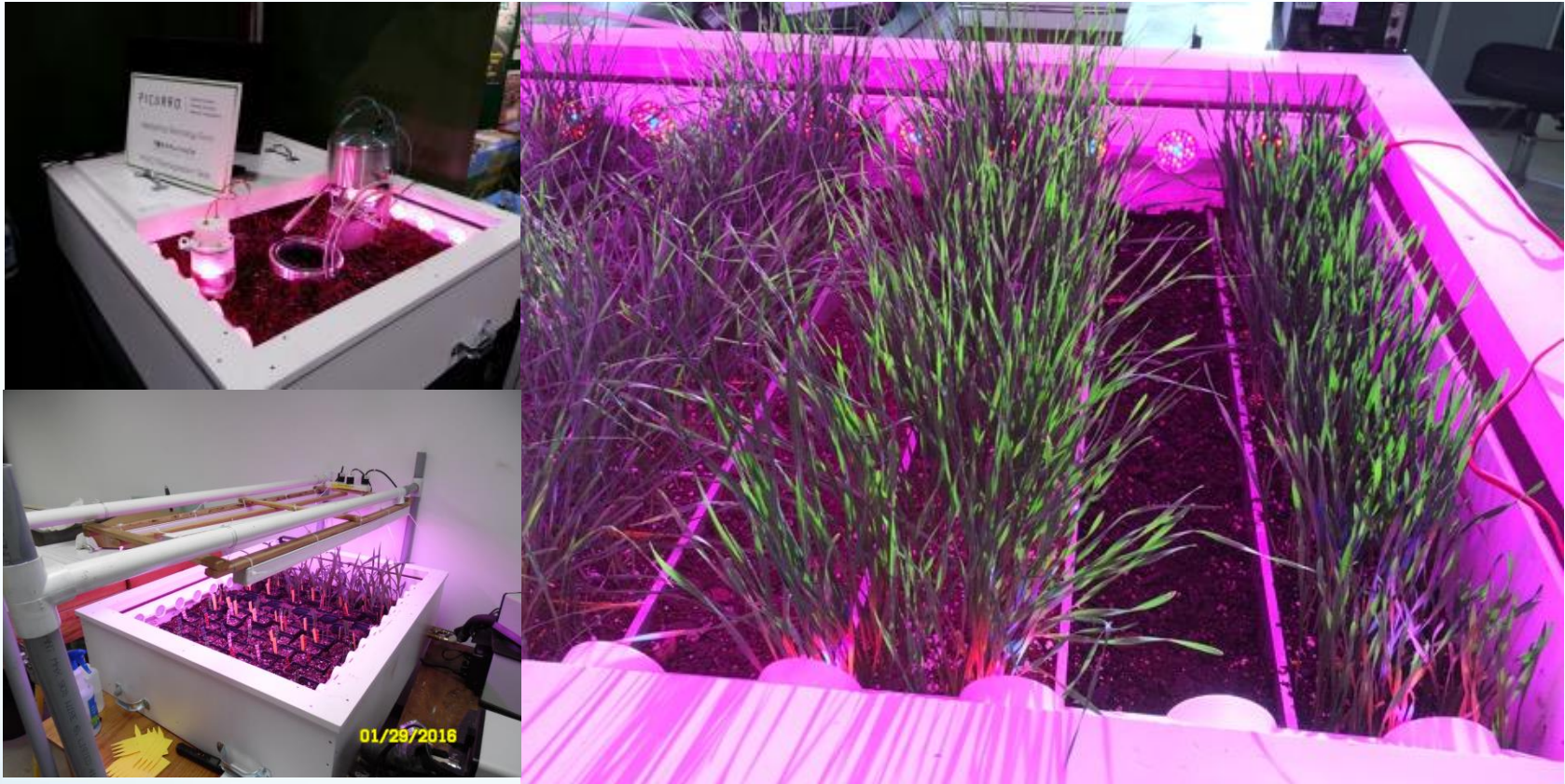
MARYLAND PLANT NUTRIENT RECOMMENDATIONS BASED ON SOIL TESTS FOR VEGETABLE CROP PRODUCTION

- For each vegetable grown in the Chesapeake Bay watershed the state department of agriculture provides specific nutrient application recommendations to be followed with the following guidance:
- For most vegetables grown on light-textured soils, apply the total recommended P_2O_5 and K_2O together with 25 to 50 percent of the recommended nitrogen before planting. The remaining nitrogen can be side dressed with a fertilizer containing nitrogen only. Side dressing or topdressing potash (K_2O) is recommended only on extremely light sandy soils with very low cation exchange capacities.
- It may be desirable to build up the phosphorus and potassium levels in very low-fertility loam and silt loam soils more rapidly than provided by these recommendations. In such instances, add an additional 40 to 50 pounds of P_2O_5 and K_2O , respectively, to the recommendations listed in the table for soils testing low in phosphorus and potassium. Apply the additional amounts in broadcast and plow down or broadcast and disk-in application.

MARYLAND PLANT NUTRIENT RECOMMENDATIONS BASED ON SOIL TESTS FOR VEGETABLE CROP PRODUCTION

- In absence of soil tests, use recommendations listed under medium phosphorus and medium potassium levels on light-textured soils that have been in intensive vegetable production.

Lab Field™ (soil environmental control system)



Video at: <https://www.jove.com/video/54647/a-gusseted-thermogradient-table-to-control-soil-temperatures-for>

Acknowledgments

- Chun-Li Wang
- Zhengxing Shen
- Jonathan Watkinson
- Novozymes North America for providing technical assistance and financial support
- Reese Jones

My family in China

